· Application No.: 09/890,478

Response to Office Action of January 28, 2005

Attorney Docket: NOTAR-005US

## Amendments to the Claims:

- 1. (currently amended) An amperometric biosensor system for the detection of analytes comprising:
  - a) at least one biocatalyst producing a pH change by its interaction with the analyte; the biocatalyst not belonging to a group of oxidoreductase enzymes;
  - b) at least one compound, in the form of a monomer, exhibiting different redox properties in its protonated and non-protonated forms (pH-sensitive redox compounds) selected from the group consisting of cyclic hydrocarbons, containing from 4 to 30 carbon atoms and substituted with at least one group selected from -OH, -SH, -NH<sub>2</sub>, =O, =S, =NH, -OR<sub>1</sub>, -SR<sub>1</sub>, -NHR<sub>1</sub>, -NR<sub>1</sub>R<sub>2</sub>, and =NR<sub>1</sub>, wherein R<sub>1</sub> and R<sub>2</sub> are hydrocarbon chains optionally further substituted, or selected from the group consisting of heterocyclic compounds containing from 3 to 30 carbon atoms and one or more heteratoms selected from the group consisting of N, S, O. Se, Te, B, P, As, Sb, and Si, optionally substituted with a group selected for -OH, -SH, -NH<sub>2</sub>, =O, =S, -NH, -OR, -SR<sub>1</sub>, -NHR<sub>1</sub>, -NR<sub>1</sub>R<sub>2</sub>, and =NR<sub>1</sub>, wherein R<sub>1</sub> and R<sub>2</sub> are independent hydrocarbon chains;
    - c) a working electrode; and
    - d) a reference electrode;
    - e) the electrodes being connected through an ammeter.
- 2. (previously presented) The biosensor system according to Claim 1, wherein the biocatalyst is selected from the group consisting of enzymes, synzymes, cells, cell components, tissues, imunoproteins, nucleic acids and extracts, fractions, fragments, homogenates, and lysates thereof.
- 3. (previously presented) The biosensor system according to Claim 2, wherein the enzyme is selected from the group consisting of hydrolase, transferase, lyase, and ligase.
- 4. (previously presented) The biosensor system according to Claim 2, wherein the enzyme is selected from the group consisting of phosophorylase, decarboxylase, esterase, phosphatase, and deaminase.

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- 5. (previously presented) The biosensor system according to Claim 2, wherein the enzyme is selected from the group consisting of urease, oxalacetate decarboxylase, glucose oxidase, carbonic anhydrase, penicillinase, and apyrase.
  - 6. (cancelled)
- 7. (currently amended) The biosensor system according to <u>Claim 1</u>, <u>Claim 6</u>, wherein the pH-sensitive redox compound is a pH indicator.
- 8. (currently amended) The biosensor system according to <u>Claim 1</u>, <u>Claim 6</u>, wherein the pH-sensitive redox compound is selected from the group consisting of hematoxylin, hematein, methylene blue, quercitin, flavonoids, alkyl gallates, polymerirthophenylenediamine and para-phenylendiamine.
- 9. (previously presented) The biosensor system according to Claim 1, wherein the working electrode is a solid composite electrode, platinum electrode, gold electrode, mercury electrode or glassy carbon electrode.
- 10. (previously presented) The biosensor system according to Claim 1, wherein the reference electrode is selected from the group consisting of Ag/AgCl and calomel electrodes.
  - 11. (cancelled)
  - 12. (cancelled)
  - 13. (cancelled)
  - 14. (previously presented) A method for detecting analytes consisting in:
    - (a) providing an amperometric biosensor system comprising:
    - (i) at least one biocatalyst producing a pH change by its interaction with the analyte; the biocatalyst not belonging to a group of oxidoreductase enzymes; the biocatalyst being inhibited by the analyte;
    - (ii) at least one compound exhibiting different redox properties in its protonated and non-protonated forms (pH-sensitive redox compounds) selected from the group consisting of cyclic hydrocarbons, containing from 4 to 30 carbon atoms and substituted with at least one group selected from -OH, -SH, -NH<sub>2</sub>, =O, =S, =NH, -OR<sub>1</sub>, -SR<sub>1</sub>, -NHR<sub>1</sub>, -NR<sub>1</sub>R<sub>2</sub>, and =NR<sub>1</sub>, wherein R<sub>1</sub> and R<sub>2</sub> are hydrocarbon chains optionally further substituted, or selected from

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the group consisting of heterocyclic compounds containing from 3 to 30 carbon atoms and one or more heteratoms selected from in the group consisting of N, S, O. Se, Te, B, P, As, Sb, and Si, optionally substituted with a group selected for -OH, -SH, -NH<sub>2</sub>, =O, =S, -NH, -OR, -SR<sub>1</sub>, -NHR<sub>1</sub>, -NR<sub>1</sub>R<sub>2</sub>, and =NR<sub>1</sub>, wherein R<sub>1</sub> and R<sub>2</sub> are independent hydrocarbon chains;

- (iii) a working electrode; and
- (iv) a reference electrode;
- (v) the electrodes being connected through an ammeter;
- (b) placing the electrodes in a measuring solution;
- (c) applying a suitable potential between the electrodes;
- (d) adding the substrate of the said biocatalyst to the measuring solution;
- (e) measuring a background current;
- (f) adding to the solution the sample containing the inhibiting-analyte to be determined;
- (g) measuring a current change that is proportional to the inhibiting-analyte concentration; and
- (h) optionally subtracting the current change measured with a blank electrode from the value obtained in (g).
- 15. (previously presented) A method for detecting analytes consisting in:
  - (a) providing an amperometric biosensor system comprising:
  - (i) at least one biocatalyst producing a pH change by its interaction with the analyte; the biocatalyst not belonging to a group of oxidoreductase enzymes; the biocatalyst being inhibited by the analyte;
  - (ii) at least one compound exhibiting different redox properties in its protonated and non-protonated forms (pH-sensitive redox compounds) selected from the group consisting of cyclic hydrocarbons, containing from 4 to 30 carbon atoms and substituted with at least one group selected from -OH, -SH, -NH<sub>2</sub>, =O, =S, =NH, -OR<sub>1</sub>, -SR<sub>1</sub>, -NHR<sub>1</sub>, -NR<sub>1</sub>R<sub>2</sub>, and =NR<sub>1</sub>, wherein R<sub>1</sub> and R<sub>2</sub> are hydrocarbon chains optionally further substituted, or selected from the group consisting of heterocyclic compounds containing from 3 to 30

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carbon atoms and one or more heteratoms selected from in-the group consisting of N, S, O. Se, Te, B, P, As, Sb, and Si, optionally substituted with a group selected for -OH, -SH, -NH<sub>2</sub>, =O, =S, -NH, -OR, -SR<sub>1</sub>, -NHR<sub>1</sub>, -NR<sub>1</sub>R<sub>2</sub>, and =NR<sub>1</sub>, wherein R<sub>1</sub> and R<sub>2</sub> are independent hydrocarbon chains;

- (iii) a working electrode; and
- (iv) a reference electrode;
- (v) the electrodes being connected through an ammeter;
- (b) applying a suitable potential between the electrodes;
- (c) adding the substrate of the said biocatalyst;
- (d) measuring a background current;
- (e) contacting the biosensor with the sample containing the inhibiting-analyte system;
- (f) measuring a current change that is proportional to the inhibitinganalyte concentration; and
- (g) optionally subtracting the current change measured with a blank electrode from the value obtained in (f).
- 16. (Cancelled)
- 17. (Previously presented) The biosensor system according to Claim 7, wherein the pH indicator is selected from the group consisting of phenoxazines, phenothizines dyes, and natural antioxidants.